

# SDH and COX activity staining in the ovary

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 An abbreviated version of this protocol was published in eLIFE in Oct 2019

Electron transport chain biogenesis activated by a JNK-insulin-Myc relay primes mitochondrial inheritance in *Drosophila*

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## Detailed protocol

### COX staining solution:

50 mM phosphate (pH 7.4),  
4 mM 3,3'-diaminobenzidine,  
2 µg/ml catalase,  
200 µM cytochrome c,  
84 mM malonate,  
60 µM rotenone,  
and 4 mM antimycin A.

### SDH staining solution:

50 mM phosphate (pH 7.4),  
42 mM succinic acid,  
0.4 mM phenazine methosulfate,  
0.5 mM nitro blue tetrazolium,  
4.5 mM EDTA,  
60 µM rotenone,  
4 mM antimycin A,  
and 2 mM KCN.

### COX inhibitor solution:

COX staining solution + 2 mM KCN

### SDH inhibitor solution:

SDH staining solution + 84 mM malonate

### Procedure (at room temperature):

#### COX/SDH dual staining: skip steps 3 to 5 for single COX staining

1. COX staining solution incubation for 30 min,
2. 3 x 5 min washes in 50 mM phosphate (pH 7.4),
3. SDH staining solution incubation for 10 min,
4. 2 x 5 min washes in 50 mM phosphate (pH 7.4),
5. 2 x 5 min washes in 50 mM phosphate (pH 7.4),
6. incubation in 80% glycerol in 50 mM phosphate (pH 7.4).

The negative controls using **COX inhibitor solution** and **SDH inhibitor solution** were performed with each batch of staining. Brightfield germarium images were collected by Zeiss Axio Observer Z1 microscope.

**How to cite:** (Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Xu, H. (2019). SDH and COX activity staining in the ovary. Bio-protocol Preprint. [bio-protocol.org/prep81](https://doi.org/10.21203/rs.3.rs-311111/v1).
2. Wang, Z., Liu, Y., Chaitankar, V., Pirooznia, M. and Xu, H. (2019). Electron transport chain biogenesis activated by a JNK-insulin-Myc relay primes mitochondrial inheritance in *Drosophila*. eLIFE. DOI: [10.7554/eLife.49309](https://doi.org/10.7554/eLife.49309)

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